

Isothermal Freezer

TEMPERATURE
UNIFORMITY TESTING



In life science research, storing materials at low temperatures has been proven to extend the viability of preserved samples.

While many store samples in mechanical freezers near the -80°C mark, it is important to note that at this temperature, metabolic activity has not ceased, merely slowed down. Reducing sample temperatures below the glass transition temperature of water (-132°C) pauses all metabolic activity. This discovery introduced liquid nitrogen (LN2) into the research lab because when in liquid state, it measures -196°C .

➤ Sample storage

Sample storage temperatures within modern LN2 cryogenic freezers will range between -150°C and -196°C , depending on the laboratory ambient environment, the distance between the freezer and the liquid nitrogen supply tank, and how the cold surrounds the sample – full LN2 submersion versus vapor storage.

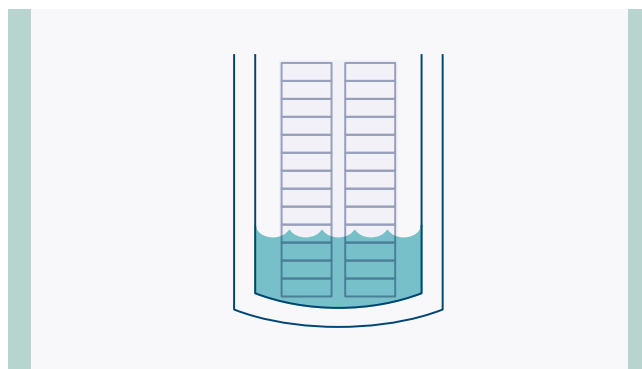


Figure 1. Sample submersion in LN2 freezer.

Over the years, submerging samples in LN2 has uncovered several risks, including sample cross-contamination, laboratory staff burns, and sample storage vessel rupture. To better account for these risks, LN2 freezer manufacturers shifted efforts to design vapor phase storage freezers where the sample should not contact LN2 directly, just benefit from the protection of the liquid nitrogen off-gassing. However, vapor phase storage has presented its own challenges, such as maintaining uniform sample temperatures based on where the sample sits within the freezer.



Figure 2. Vapor storage with perforated base supporting samples above LN2.

CBS Cryogenic Solutions, part of BioLife Solutions, designed and initially patented an Isothermal Cryogenic Freezer that insulates the inner sample chamber with a jacket of liquid nitrogen. The freezer is designed to off-gas the LN2 vapor into the sample storage chamber – surrounding the samples in true LN2 vapor while also using the liquid in the jacket to further promote temperature uniformity throughout the tank.



Figure 3. Isothermal freezer jacketed LN2 design providing true vapor storage.

With any industry-first, the concept must be scientifically proven, and this white paper provides supporting evidence that the V-Series Isothermal Freezers provide true dry-vapor sample protection with tank uniformity that will maximize sample storage within the freezer.

➤ METHOD

For this test, the CBS Cryogenic Solutions V-1500 freezer model was tested with a Kaye Validator and nine (9) temperature probes over a 24-hour period. The temperature probes were mapped according to **Figure 4** to best measure ambient temperature and various spots within the freezer.

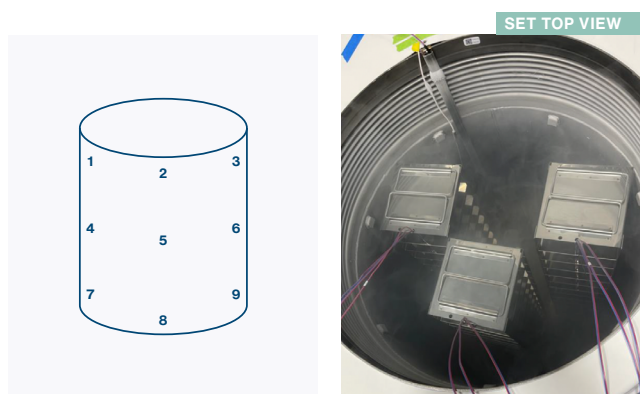


Figure 4. Temperature probe locations during test.

Probes 1,2,3 are placed in the “top box” configuration within the rack. They measure 2.5 inches below the lid of the freezer. Probes 4,5,6 are referencing the “middle box” location and they are 17.5 inches below the lid. Probes 7,8,9 represent the “bottom box” in the sample rack system, and they measure 28.5 inches below the lid.

A 24-hour test period was run to measure the freezer temperatures between automatic LN2 refills. The freezer was filled with three aluminum, standard two-inch box racks (part number 1201APLR-O) to hold the temperature probes in place. This test configuration has become the IQ/OQ testing standard used for infield customer validations.

Results

Every five (5) minutes, temperature measurements were recorded from each temperature probe. These temperature measurements were averaged each hour and are listed in Table 1 by probe number.

| Hour | Probe 1 | Probe 2 | Probe 3 | Probe 4 | Probe 5 | Probe 6 | Probe 7 | Probe 8 | Probe 9 |
|---------------|---------|---------|---------|---------|---------|---------|---------|---------|---------|
| 1 | -189.4 | -188.9 | -189.4 | -194.6 | -194.7 | -194.7 | -195.2 | -195.2 | -195.2 |
| 2 | -188.0 | -187.4 | -188.0 | -194.3 | -194.4 | -194.4 | -195.0 | -195.0 | -195.0 |
| 3 | -187.0 | -186.3 | -186.9 | -194.0 | -194.3 | -194.2 | -194.9 | -194.9 | -194.9 |
| 4 | -186.3 | -185.6 | -186.3 | -193.9 | -194.1 | -194.0 | -194.9 | -194.9 | -194.9 |
| 5 | -185.9 | -185.2 | -185.9 | -193.8 | -194.0 | -194.0 | -194.9 | -194.8 | -194.9 |
| 6 | -185.6 | -184.9 | -185.6 | -193.7 | -194.0 | -193.9 | -194.8 | -194.8 | -194.9 |
| 7 | -185.4 | -184.7 | -185.4 | -193.6 | -193.9 | -193.8 | -194.8 | -194.8 | -194.8 |
| 8 | -185.2 | -184.5 | -185.2 | -193.5 | -193.8 | -193.7 | -194.8 | -194.8 | -194.8 |
| 9 | -185.0 | -184.3 | -185.0 | -193.4 | -193.8 | -193.7 | -194.8 | -194.8 | -194.8 |
| 10 | -184.9 | -184.2 | -184.8 | -193.3 | -193.7 | -193.6 | -194.8 | -194.8 | -194.8 |
| 11 | -184.7 | -184.0 | -184.6 | -193.2 | -193.6 | -193.5 | -194.8 | -194.8 | -194.8 |
| 12 | -184.5 | -183.8 | -184.4 | -193.1 | -193.5 | -193.4 | -194.8 | -194.8 | -194.8 |
| 13 | -184.3 | -183.6 | -184.3 | -192.9 | -193.4 | -193.3 | -194.8 | -194.7 | -194.8 |
| 14 | -184.1 | -183.5 | -184.1 | -192.8 | -193.3 | -193.2 | -194.8 | -194.7 | -194.8 |
| 15 | -184.0 | -183.3 | -183.9 | -192.7 | -193.2 | -193.1 | -194.7 | -194.7 | -194.7 |
| 16 | -183.8 | -183.1 | -183.7 | -192.5 | -193.1 | -192.9 | -194.7 | -194.7 | -194.7 |
| 17 | -183.6 | -182.9 | -183.6 | -192.4 | -193.0 | -192.8 | -194.7 | -194.7 | -194.7 |
| 18 | -183.4 | -182.7 | -183.4 | -192.2 | -192.8 | -192.7 | -194.7 | -194.7 | -194.7 |
| 19 | -183.3 | -182.5 | -183.2 | -192.0 | -192.7 | -192.5 | -194.7 | -194.6 | -194.7 |
| 20 | -183.1 | -182.3 | -183.0 | -191.9 | -192.6 | -192.4 | -194.6 | -194.6 | -194.7 |
| 21 | -182.9 | -182.1 | -182.8 | -191.7 | -192.4 | -192.3 | -194.6 | -194.6 | -194.7 |
| 22 | -182.7 | -181.9 | -182.6 | -191.5 | -192.3 | -192.1 | -194.6 | -194.6 | -194.6 |
| 23 | -182.5 | -181.7 | -182.4 | -191.3 | -192.1 | -191.9 | -194.6 | -194.6 | -194.6 |
| 24 | -182.3 | -181.5 | -182.2 | -191.2 | -191.9 | -191.7 | -194.6 | -194.6 | -194.6 |
| 24-Hr Average | -184.7 | -183.9 | -184.6 | -192.9 | -193.4 | -193.2 | -194.8 | -194.8 | -194.8 |

Table 1. Averaged hourly temperature recordings by probe.

All averaged hourly temperature measurements were then plotted over the 24-hour period, by probe number. This graph is shown in **Figure 5**.

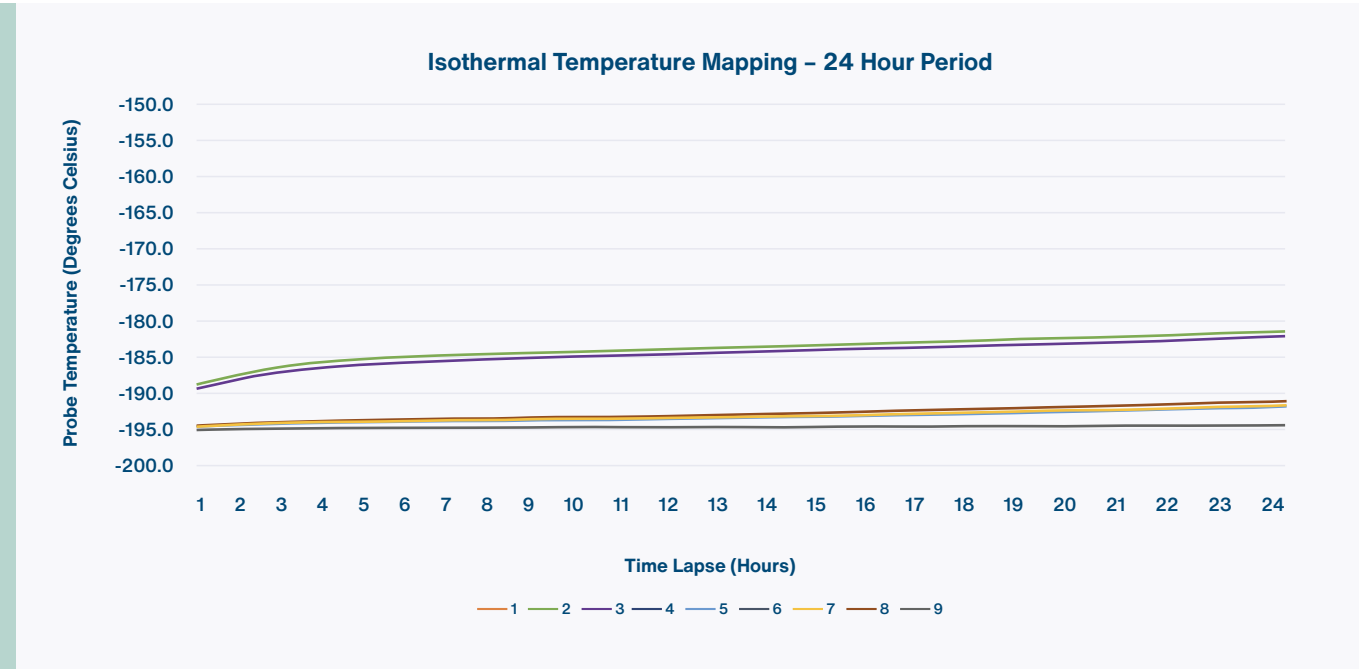


Figure 5. Averaged hourly temperature plots by probe.

The average line of all probes is plotted below in **Figure 6**.

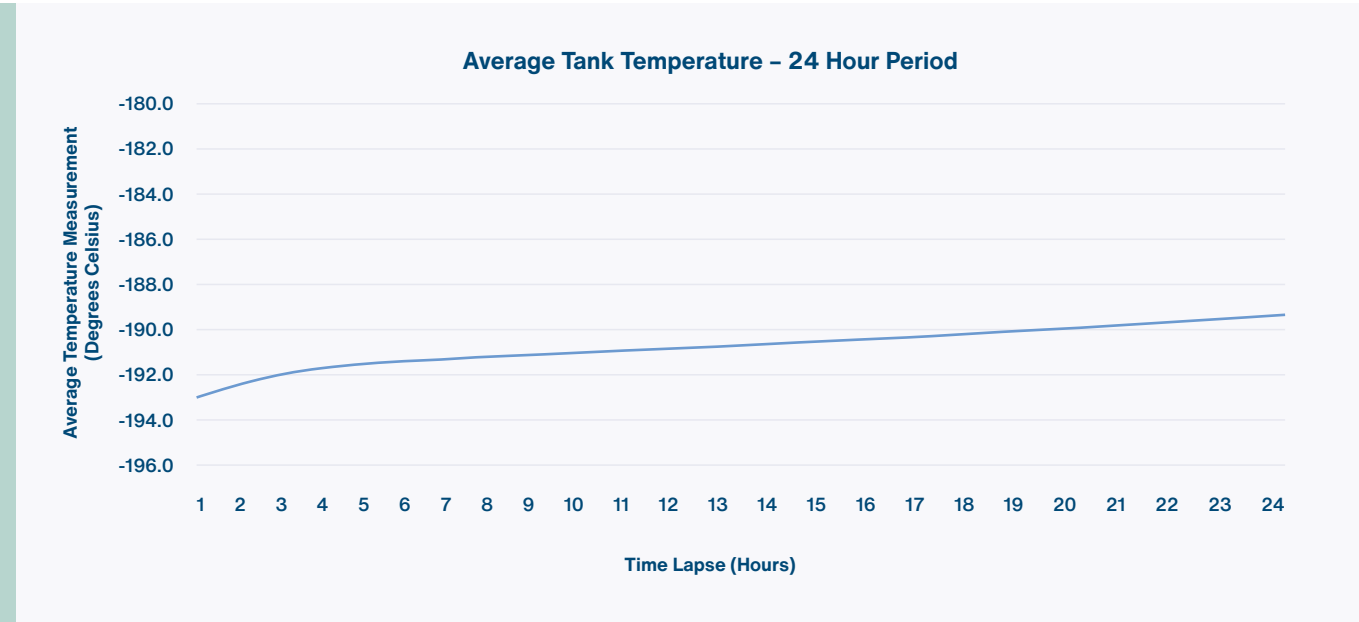


Figure 6. Average freezer temperature over a 24-hour period.

The average freezer temperature over a 24-hour period sits at -190°C, with a $\pm 3^{\circ}\text{C}$ variation. This temperature uniformity profile has been averaged from the top of the freezer to the bottom.

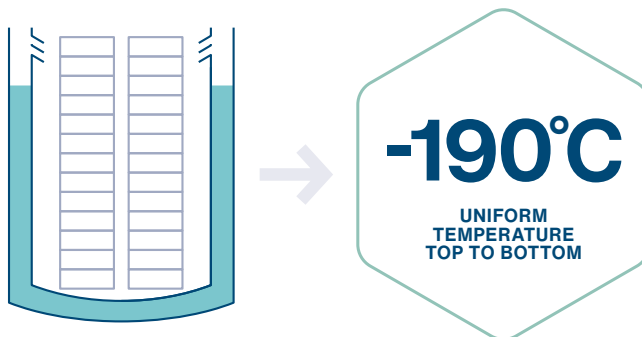
➤ Conclusion

The CBS Cryogenic Solutions V-Series Isothermal Freezers provide uniform protection for samples, well below the glass transition temperature of water (-132°C). This test was conducted with a nearly empty chamber, aside from the three aluminum racks, and can therefore be considered a worst-case scenario for sample temperature measurements. When samples are stored inside these freezers, they hold an added thermal mass to further stabilize the temperature uniformity within the freezer.

By design, samples stored within an Isothermal freezer will never encounter liquid nitrogen – leaving the entire tank volume to fill with samples. Most LN2 vapor storage freezers currently available on the market only offer the space above the LN2 storage area and state a sample temperature range of -150°C to -190°C in their literature

with limited evidence to verify the freezer temperature profile from top of the tank (furthest from LN2) to the bottom (closest to LN2).

-132°C is the absolute warmest a biological sample can be to stop metabolic activity. To ensure your samples stay below this temperature and have no direct contact with LN2, choose a V-Series Isothermal freezer.



Isothermal LN2 Freezers

Innovative LN2 Jacket Technology

